

Project No. _____

Book No. _____

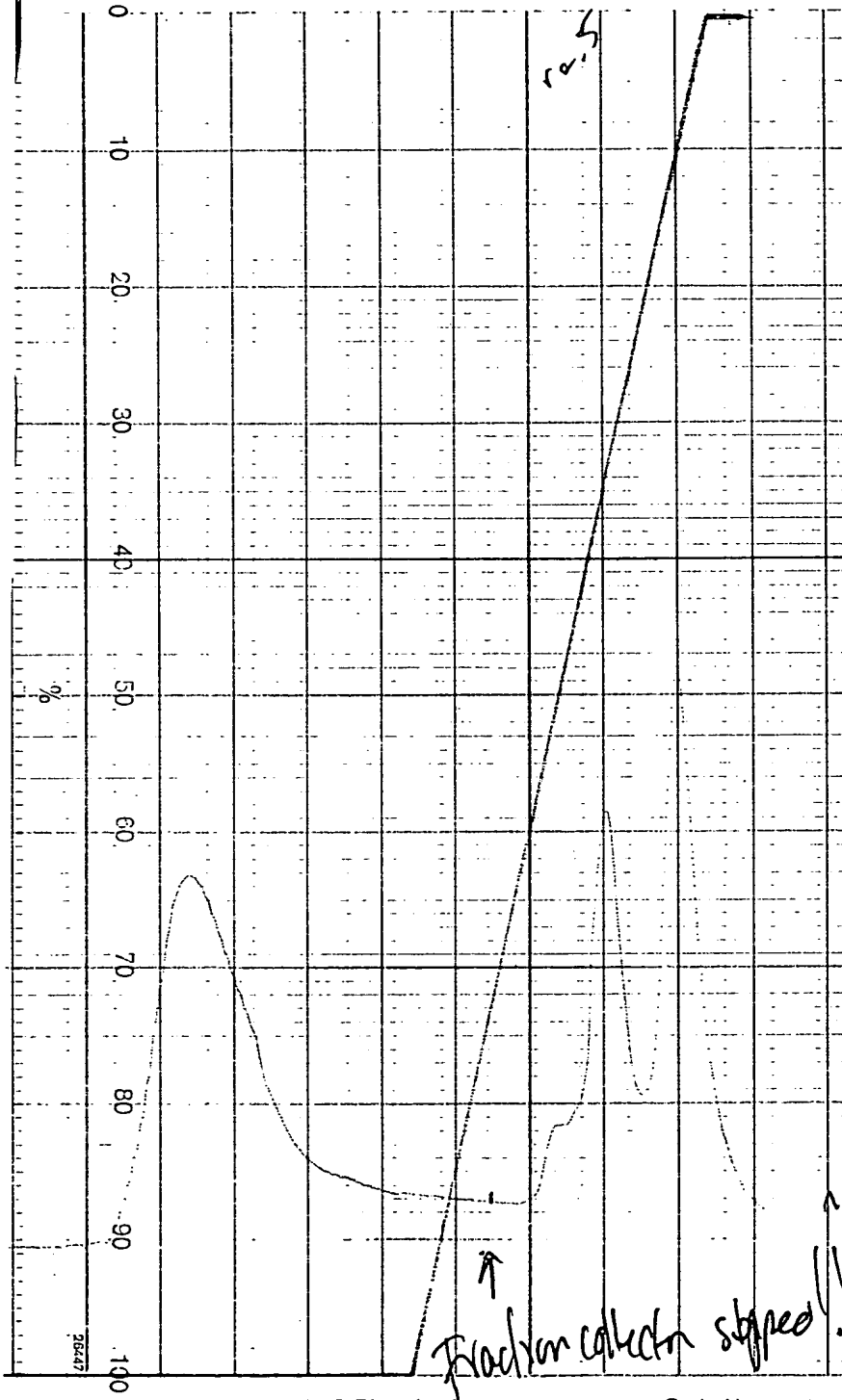
TITLE

FY-1 Q6SDM-2mL

146

From Page No. _____

06/1



Q Buffer A -

25mM Phos - pH 7.2
1mM EDTA
10mM KCl
5mM Bme
10% glycerol

Q Buffer B

25mM Phos - pH 7.2
1mM EDTA
800mM KCl
5mM Bme
10% glycerol

6/20/95

Pharmacia LKB Biotechnology

Code No. 18-10C

To Page 1

Witnessed & Understood by me,

Date

Invented by

Date

May Longo

6/20/95

Recorded by

S. Hyman

06/16/95

Tag No. _____

06/16

Bump + column w/ .5N NaOH -

* FY-1

wash extensively w/ H₂O

equilibrate w/ Q650-Buffer A - p. 146 -

Load ~ 3.5 mL of Heparin pool of FY-1 @ .5 mL/min

Wash with QBuffer A until base line is reached -

Gradient - 20 mL linear gradient 0-100% Qbuffer B
@ .5 mL/min collect .5 mL fractionsWash w/ 10 mL of 100% Qbuffer B - collect
.5 mL fraction fractions -

Fraction collector - started then stopped after
fraction 10!! - Did not realize until
gradient was finished - lost entire elution
~~elution~~ to waste! Could have tried to
save however I believe I washed the port
with .2N NaOH + in the same waste container.

Fraction collector stopped b/c outside of rack was "dirty"
and was slippery - Must be sure outside plastic is
clean!

Can to proceed with 3'-5' exo mutant - Bump column
with 3M KCl - wash w/ H₂O equilibrate w/
Q650 Buffer A p. 146.

To Page No. _____

Read & Understood by me,

Date

Invented by

Date

May Jones

6/20/95

Recorded by

06/14/95

Project No. _____

Book No. _____

TITLE

Q650M - 3'5' x 0 minus - T₁

From Page No. _____

4/1

0-800 mM KCl in @ pH 7.2

0806, N50/99109/8.93

SAM	CFM1
1	20 188.00
2	22 138.00
3	24 180.00
4	26 874.00
5	28 830.00
6	30 748.00
7	32 1174.00
8	34 912.00
9	36 556.00
10	38 590.00
11	40 340.00
12	42 326.00
13	44 370.00
14	46 266.00
15	48 298.00
16	50 186.00
17	9928.00
18	198.00

Fracture

Pool

Pool 26-3
dialyze of
in JFAQ
storage

Pool 26-35

6/20/95

6/11/95

iotechnology

Code No. 18-1001-44

T Page N

Witnessed & Understood by me,

Date

Invented by

Dat

Man Jones

6/20/95

Rec rded by

6/12/95

Tag No. _____

Equilibrate 2mL Q650 m w/ Q buffer A - 6/17
 dilute load of 3

Load @ .5 mL/min - ~~Sens~~ Sensitivity - .05

Wash to base w/ Buffer A - collect F.T. - @ 1 mL/min

Program - ① 5mL Wash w/ Q Buffer A @ .5 mL/min
 ② 20mL linear gradient 0 → 100% Q Buffer B
 @ .5 mL/min
 ③ 4.5mL Wash w/ Q Buffer B @ .5 mL/min
 collect 500 μ L fractions -

Assay -

μ L of premix aliquotted to prelabeled ependorfs -
 nuclease @ 74°C for 5 min quench w/
 10 μ L of .5M EDTA - spot 20 μ L on 6 PIC
 filters - TCA wash

1x 10% TCA + 1% Pi @ 5'

3x 5% TCA + @ 5'

2x EtOH

dry + count in LSF - Econofluor

Pool 2u-35 dialyze o/N (over weekend) - against
 TAE storage buffers (No detergents) -

? - Remove ~ 1.8mL from dialysis - store in 2mL eppendorf
 HOT PINK - -20°C

To Page No. _____

Read & Understood by m ,

Date

Invented by

Date

Way Longo

6/20/95

Recorded by

S. H. m

6/19/95